

Claims:

1. A chimeric antibody conjugate comprising an antigen binding region of a non-human antibody and an immunoglobulin constant region which  
 5 comprises at least one C<sub>H</sub> domain ~~or epitope thereof~~, with the proviso that the constant region is not a naturally occurring F<sub>C</sub> fragment.
2. A chimeric antibody conjugate according to claim 1 in which the non-human antigen binding region comprises or consists of a non-human Fab  
 10 fragment or part thereof.
3. A chimeric antibody conjugate according to <sup>claim 1</sup> ~~claim 1 or claim 2~~ in which the non-human antigen binding region comprises or consist of an scFv fragment.  
 15
4. A chimeric antibody conjugate according to <sup>claim 1</sup> ~~any one of claims 1 to 3~~ in which the non-human antigen binding region is derived from a mouse.
5. A chimeric ~~antibody~~ conjugate according to <sup>claim 1</sup> ~~any one of claims 1 to 4~~ in which the constant region is derived from a human antibody.  
 20
6. A chimeric ~~antibody~~ conjugate according to <sup>claim 1</sup> ~~any one of claims 1 to 5~~ in which the constant region comprises one or more constant domains derived from an IgM antibody.  
 25
7. A chimeric ~~antibody~~ conjugate according to claim 6 in which the constant region **comprises one** or more C<sub>H</sub>3μ domains.
8. A chimeric ~~antibody~~ conjugate according to <sup>claim 1</sup> ~~any one of claims 1 to 5~~ in which the constant region comprises one or more constant domains derived from an IgG antibody.  
 30
9. A chimeric antibody conjugate according to claim 8 in which the constant region comprises one or more C<sub>H</sub>3γ domains.  
 35

*a* 10. A chimeric antibody conjugate according to <sup>Claim 1</sup>~~any one of claims 1 to 5~~ in which the constant region comprises one or more constant domains derived from an IgA antibody.

*a* 5 11. A chimeric antibody conjugate according to <sup>Claim 1</sup>~~any one of claims 1 to 10~~ in which the constant region comprises a non-naturally occurring combination of C<sub>H</sub> domains or epitopes thereof.

*a* 10 12. A chimeric antibody conjugate according to <sup>Claim 1</sup>~~any one of claims 1 to 11~~ in which the non-human antigen binding region binds to an epitope of an infectious agent selected from dengue virus, rubella virus, herpes virus, parvovirus, human glycoporphin, *Rickettsia sibirica*, *Burkholderia pseudomallei*, *Salmonella typhi* or *paratyphi*, *Leptospira interrogans*, *Plasmodium falciparum/vivax*, Japanese encephalitis virus, Yellow fever virus, *Bordetella pertussis/parapertussis*, *Candida albicans/kruzei*, Varicella zoster virus, HIV, 15 Hepatitis viruses, Human papilloma virus, Epstein-Barr virus, Ross River virus, *Brucella abortis*, Human herpesvirus-6, Parvovirus B19, *Coxiella burnettii*, Herpes simplex viruses 1&2, *Rickettsia rickettsii*, *Conori australis*, and *Rickettsia tsutsugamushi*.

20 13. A recombinant polynucleotide molecule comprising a sequence encoding a non-human V<sub>H</sub> region, a sequence encoding a non-human V<sub>L</sub> region, a sequence encoding a flexible linker positioned between the V<sub>H</sub> region sequence and the V<sub>L</sub> region sequence, and a heterologous sequence 25 encoding a C<sub>H</sub> domain or epitope thereof.

14. A recombinant polynucleotide molecule according to claim 13 in which the heterologous sequence encodes a human C<sub>H</sub> domain.

*a* 30 15. A recombinant polynucleotide molecule according to claim 13 ~~or claim 14~~ in which the C<sub>H</sub> domain sequence is linked to the 3' end of the V<sub>L</sub> or V<sub>H</sub> sequence.

*a* 35 16. A recombinant polynucleotide molecule according to <sup>Claim 13</sup>~~any one of claims 13 to 15~~ in which the polynucleotide molecule includes a control

sequence which directs the synthesis of both the V<sub>L</sub> and V<sub>H</sub> polypeptide regions.

17. A recombinant polynucleotide molecule according to claim 16 in which the control sequence is the lac promoter.

18. A recombinant polynucleotide molecule according to <sup>claim 13</sup> ~~any one of claims 13 to 17~~ in which the molecule includes a sequence encoding a leader peptide which directs the synthesised polypeptide chains to the host cell periplasm.

19. A recombinant polynucleotide molecule according to claim 18 in which the leader sequence is the pel B sequence.

20. A recombinant polynucleotide molecule comprising a sequence encoding a non-human V<sub>L</sub> region, a sequence encoding a non-human C<sub>L</sub> region, a sequence encoding a non-human V<sub>H</sub> region, a heterologous sequence encoding a C<sub>H</sub> domain or epitope thereof, and optionally a sequence encoding a non-human C<sub>H1</sub> region.

21. A recombinant polynucleotide molecule according to claim 21 in which the heterologous sequence encodes a human C<sub>H</sub> domain.

22. A recombinant polynucleotide molecule according to claim 20 ~~or claim 21~~ in which the V<sub>L</sub> and C<sub>L</sub> sequences are linked together so that the V<sub>L</sub> and C<sub>L</sub> regions are expressed as a single polypeptide.

23. A recombinant polynucleotide molecule according to <sup>claim 20</sup> ~~any one of claims 20 to 22~~ in which the V<sub>H</sub> and C<sub>H1</sub> sequences are linked together so that the V<sub>H</sub> and C<sub>H1</sub> regions are expressed as a single polypeptide.

24. A recombinant polynucleotide molecule according to <sup>claim 20</sup> ~~any one of claims 20 to 23~~ in which the polynucleotide molecule includes a control sequence which directs the synthesis of both the V<sub>L</sub>-C<sub>L</sub> and V<sub>H</sub>-C<sub>H1</sub> polypeptide regions.

25. A recombinant polynucleotide molecule according to claim 24 in which the control sequence is the lac promoter.

a 26. A recombinant polynucleotide molecule according to <sup>Claim 20</sup> ~~any one of~~  
5 ~~claims 20 to 25~~ in which the polynucleotide molecule includes a sequence encoding a leader peptide which directs the synthesised polypeptide chains to the host cell periplasm.

27. A recombinant polynucleotide molecule according to claim 26 in  
10 which the leader sequence is the pel B sequence.

a 28. A recombinant polynucleotide molecule according to <sup>Claim 20</sup> ~~any one of~~  
a ~~claims 20 to 27~~ in which the heterologous C<sub>H</sub> domain sequence is linked to  
15 the V<sub>L</sub>-C<sub>L</sub> sequences or the V<sub>H</sub>-C<sub>H</sub>1 sequences so that the expressed heterologous C<sub>H</sub> domain is attached to the V<sub>L</sub>-C<sub>L</sub> polypeptide or the V<sub>H</sub>-C<sub>H</sub>1 polypeptide.

a 29. A vector comprising a polynucleotide according to <sup>Claim 13</sup> ~~any one of claims~~  
a ~~13 to 29~~.

20 30. A bacterial, yeast, insect or mammalian host cell transformed with a vector according to claim 29.

25 31. A method of producing a chimeric antibody conjugate which comprises culturing a host cell according to claim 30 under conditions enabling the expression of the conjugate and optionally recovering the conjugate.

30 32. A chimeric antibody conjugate produced by a method according to claim 31.

a 33. A method for detecting an antibody in a biological sample which  
a involves comparing the level of detection obtained with the biological sample to the level of detection obtained with a positive control, wherein the  
35 positive control comprises a chimeric antibody conjugate according to <sup>Claim 1</sup> ~~any one of claims 1 to 12~~.

34. A method according to claim 33 in which the biological sample is a human biological sample.

a 5 35. A method according to claim 33 ~~or claim 34~~ in which the antibodies to be detected are antibodies characteristic of a disease selected from dengue fever, japanese encephalitis, rubella, spotted fever, herpes infection, parvovirus infections, melioidosis, typhoid, leptospirosis, malaria, yellow fever, whooping cough, systemic candidiasis/thrush, chicken pox, shingles. 10 AIDS, hepatitis, liver cancer, cervical cancer, infectious mononucleosis, nasopharyngeal carcinoma, Ross River fever, brucella, exanthum subitum (sixth disease/roseola infantum), erythema infectiosum (fifth disease), Q fever, cold sores, genital herpes, spotted fever and scrub typhus.

a 15 36. A method according to <sup>Claim 33</sup> ~~any one of claims 33 to 35~~ in which the antibody is an IgM antibody.

a 37. A method according to <sup>Claim 33</sup> ~~any one of claims 33 to 35~~ in which the antibody is an IgG antibody.

20 38. A method according to <sup>Claim 33</sup> ~~any one of claims 33 to 35~~ in which the antibody is an IgA antibody.

a 39. A bifunctional molecule for use in labelling an antibody derived from 25 a first species, the bifunctional molecule comprising a binding region which binds to the antibody of the first species or to one or more groups provided thereon, and a constant region derived from an antibody of a second species, the constant region comprising at least one C<sub>H</sub> domain or an epitope thereof.

30 40. A bifunctional molecule according to claim 39 in which the binding and constant regions are separated by a linker molecule.

41. A bifunctional molecule according to claim 40 in which the linker molecule is a peptide of between 1 and 20 amino acids in length.

35

42. A bifunctional molecule according to claim 41 in which the linker molecule is a peptide of between 2 and 5 amino acids in length.

a 43. A bifunctional molecule according to <sup>Claim 39</sup> ~~any one of claims 39 to 42~~ in which the binding region is not derived from an antibody.

a 44. A bifunctional molecule according to <sup>Claim 39</sup> ~~any one of claims 39 to 43~~ in which the binding region binds directly to the antibody derived from the first species.

10 45. A bifunctional molecule according to claim 44 in which the binding region is derived from a protein selected from the group consisting of, *Streptococcal* protein G, *Staphylococcal aureus* protein A and *Peptostreptococcus magnus* protein L.

15 46. A bifunctional molecule according to claim 45 in which the binding region comprises fragment B of *Staphylococcus aureus* protein A.

20 47. A bifunctional molecule according to claim 44 in which the binding region comprises a mouse Fc  $\gamma$  receptor or fragment thereof.

48. A bifunctional molecule according to claim 44 in which the binding region comprises a histidine rich glycoprotein.

a 25 49. A bifunctional molecule according to <sup>Claim 39</sup> ~~any one of claims 39 to 43~~ in which the binding region binds to one or more groups provided on the antibody of the first species.

30 50. A bifunctional molecule according to claim 49 in which the group(s) is a biotin molecule and the binding region comprises streptavidin or a fragment thereof.

a 35 51. A bifunctional molecule according to <sup>Claim 39</sup> ~~any one of claims 39 to 50~~ in which the antibody constant region is not a naturally occurring Fc fragment.

a

52. A bifunctional molecule according to <sup>Claim 39</sup> ~~any one of claims 39 to 51~~ in which the constant region comprises one or more constant domains derived from an IgM antibody.

5 53. A bifunctional molecule according to claim 52 in which the constant region comprises one or more  $C_H3\mu$  domains.

a

54. A bifunctional molecule according to <sup>Claim 39</sup> ~~any one of claims 39 to 51~~ in which the constant region comprises one or more constant domains derived from an IgG antibody.

10

55. A bifunctional molecule according to claim 54 in which the constant region comprises one or more  $C_H3\gamma$  domains.

a

56. A bifunctional molecule according to <sup>Claim 39</sup> ~~any one of claims 39 to 51~~ in which the constant region comprises one or more constant domains derived from an IgA antibody.

15

a

57. A bifunctional molecule according to <sup>Claim 39</sup> ~~any one of claims 39 to 56~~ in which the antibody constant region comprises or consists of a non-naturally occurring combination of immunoglobulin  $C_H$  domains or epitopes thereof.

20

a

58. A bifunctional molecule according to <sup>Claim 39</sup> ~~any one of claims 39 to 56~~ in which the antibody constant region comprises or consists of a single  $C_H$  domain.

25

a

59. A bifunctional molecule according to <sup>Claim 39</sup> ~~any one of claims 39 to 58~~ in which the second species is a human.

a

60. An isolated polynucleotide encoding a bifunctional molecule according to <sup>Claim 39</sup> ~~any one of claims 39 to 59~~.

30

61. A vector comprising a polynucleotide according to claim 60.

62. A bacterial, yeast, insect or mammalian host cell transformed with a vector according to claim 61.

35

63. A method of producing a bifunctional molecule which comprises culturing a host cell according to claim 62 under conditions enabling the expression of the bifunctional molecule and optionally recovering the  
5 bifunctional molecule.

64. A bifunctional molecule produced by a method according to claim 63.

10 65. A complex formed between (i) an antibody or biologically active fragment thereof derived from a first species and (ii) a bifunctional molecule, the bifunctional molecule comprising a binding region which binds to the antibody of the first species or to one or more groups provided thereon, and a  
15 constant region derived from an antibody of a second species, the constant region comprising at least one C<sub>H</sub> domain or an epitope thereof.

66. A complex according to claim 65 in which the binding region has a K<sub>D</sub> for the antibody of the first species, or a group provided thereon, of less than 10<sup>-6</sup> M.  
20

67. A complex according to claim 66 in which the binding region has a K<sub>D</sub> for the antibody of the first species, or a group provided thereon, of less than 10<sup>-8</sup> M.

25 68. A complex according to <sup>claim 65</sup> ~~any one of claims 65 to 67~~ in which the bifunctional molecule binds directly to the antibody derived from the first species.

69. A complex according to claim 68 in which the binding region is  
30 derived from a protein selected from the group consisting of, *Streptococcal* protein G, *Staphylococcal aureus* protein A and *Peptostreptococcus magnus* protein L.

70. A complex according to claim 69 in which the binding region  
35 comprises fragment B of *Staphylococcus aureus* protein A.



71. A complex according to claim 68 in which the binding region comprises a mouse Fc  $\gamma$  receptor or fragment thereof.

5 72. A complex according to claim 68 in which the binding region comprises a histidine rich glycoprotein.

a 73. A complex according to any <sup>Claim 65</sup> ~~one of claims 65 to 67~~ in which the binding region binds to one or more groups provided on the antibody of the first species.

10

74. A complex according to claim 73 in which the group(s) is a biotin molecule and the binding region comprises streptavidin or a fragment thereof.

15

a 75. A complex according to any <sup>Claim 65</sup> ~~one of claims 65 to 74~~ in which the constant region comprises one or more constant domains derived from an IgM antibody.

20

76. A complex according to claim 75 in which the constant region comprises one or more C<sub>H</sub>3 $\mu$  domains.

25

a 77. A complex according to any <sup>Claim 65</sup> ~~one of claims 65 to 74~~ in which the constant region comprises one or more constant domains derived from an IgG antibody.

78. A complex according to claim 77 in which the constant region comprises one or more C<sub>H</sub>3 $\gamma$  domains.

30

a 79. A complex according to any <sup>Claim 65</sup> ~~one of claims 65 to 74~~ in which the constant region comprises one or more constant domains derived from an IgA antibody.

35

a 80. A complex according to any <sup>Claim 65</sup> ~~one of claims 65 to 79~~ in which the antibody constant region comprises or consists of a non-naturally occurring combination of immunoglobulin C<sub>H</sub> domains or epitopes thereof.

- a 81. A complex according to <sup>Claim 65</sup> ~~any one of claims 65 to 79~~ in which the antibody constant region comprises or consists of a single C<sub>H</sub> domain.
- a 82. A complex according to <sup>Claim 65</sup> ~~any one of claims 65 to 81~~ in which the first  
5 species is a rat or mouse.
- a 83. A complex according to <sup>Claim 65</sup> ~~any one of claims 65 to 82~~ in which the second species is a human.
- 10 84. A method for detecting an antibody in a biological sample which involves comparing the level of detection obtained with the biological sample to the level of detection obtained with a positive control, wherein the  
a positive control comprises a complex according to <sup>Claim 65</sup> ~~any one of claims 65 to 83~~.
- 15 85. A method according to claim 84 in which the biological sample is a human biological sample.
- a 86. A method according to claim 84 ~~or claim 85~~ in which the antibodies to be detected are antibodies characteristic of a disease selected from dengue  
20 fever, Japanese encephalitis, rubella, spotted fever, herpes infection, parvovirus infections, melioidosis, typhoid, leptospirosis, malaria, yellow fever, whooping cough, systemic candidiasis/thrush, chicken pox, shingles, AIDS, hepatitis, liver cancer, cervical cancer, infectious mononucleosis, nasopharyngeal carcinoma, Ross River fever, brucella, exanthum subitum  
25 (sixth disease/roseola infantum), erythema infectiosum (fifth disease), Q fever, cold sores, genital herpes, spotted fever and scrub typhus.
- a 87. A method according to <sup>Claim 84</sup> ~~any one of claims 84 to 86~~ in which the antibody is an IgM antibody.  
30
- a 88. A method according to <sup>Claim 84</sup> ~~any one of claims 84 to 86~~ in which the antibody is an IgG antibody.
- a 89. A method according to <sup>Claim 84</sup> ~~any one of claims 84 to 86~~ in which the  
35 antibody is an IgA antibody.